

Editorial comments:

Changes to be made by the Author(s):

1. Please take this opportunity to thoroughly proofread the manuscript to ensure that there are no spelling or grammar issues. The JoVE editor will not copy-edit your manuscript and any errors in the submitted revision may be present in the published version.

We have carefully revised the manuscript.

2. Please revise lines 40-44, 116-119, 210-212, 218-221, 227-231, 237-243 to avoid previously published text.

We have revised the text.

3. Please obtain explicit copyright permission to reuse any figures from a previous publication. Explicit permission can be expressed in the form of a letter from the editor or a link to the editorial policy that allows re-prints. Please upload this information as a .doc or .docx file to your Editorial Manager account. The Figure must be cited appropriately in the Figure Legend, i.e. "This figure has been modified from [citation]."

The permission has been obtained from PNAS and include in the revised material.

4. Figure 1: Please line up the panel labels better.

The figures have been corrected.

5. Figure 2: Please include a space between the number and the units of the scale bar.

The figures have been corrected.

6. Please upload each Figure individually to your Editorial Manager account as a .png, .tiff, .pdf, .svg, .eps, .psd, or .ai file.

The appropriate.ai files have been uploaded.

7. Table 1: Please upload it to your Editorial Manager account as an .xls or .xlsx file.

The appropriate.xls files have been uploaded.

8. Table of Materials: Please sort the items in alphabetical order according to the name of material/equipment.

The required changes have been made.

9. Please provide an email address for each author.

We have included emails for all the authors.

10. Please add a Summary section before the Abstract to clearly describe the protocol and its applications in complete sentences between 10-50 words: "Here, we present a protocol to ..."

We have added the summary.

11. Please define all abbreviations before use.

We have revised the text.

12. Please use SI abbreviations for all units: L, mL, μ L, h, min, s, etc. Please use the micro symbol μ instead of u. Please abbreviate liters to L to avoid confusion.

We have revised the text.

13. Please include a space between all numerical values and their corresponding units: 15 mL, 37 °C, 60 s; etc.

We have revised the text.

14. Please convert centrifuge speeds to centrifugal force (x g) instead of revolutions per minute (rpm).

We have revised the text.

15. JoVE cannot publish manuscripts containing commercial language. This includes trademark symbols ([™]), registered symbols ([®]), and company names before an instrument or reagent. Please remove all commercial language from your manuscript and use generic terms instead. All commercial products should be sufficiently referenced in the Table of Materials and Reagents. You may use the generic term followed by "(see table of materials)" to draw the readers' attention to specific commercial names. Examples of commercial sounding language in your manuscript are: The Jackson Laboratory, Thermo Fisher Scientific, Sigma, Beckman, BioLegend, BD FACScalibur, Life Technologies, Styrofoam, Zeiss AxioObserver, etc.

We have included all the information in Table of Materials.

16. Please revise the protocol text to avoid the use of any personal pronouns (e.g., "we", "you", "our" etc.).

We have revised the text.

17. Please revise the protocol to contain only action items that direct the reader to do something (e.g., “Do this,” “Ensure that,” etc.). The actions should be described in the imperative tense in complete sentences wherever possible. Avoid usage of phrases such as “could be,” “should be,” and “would be” throughout the Protocol. Any text that cannot be written in the imperative tense may be added as a “Note.” Please include all safety procedures and use of hoods, etc. However, notes should be used sparingly and actions should be described in the imperative tense wherever possible. Please move the discussion about the protocol to the Discussion.

We have revised the text.

18. Please revise the Protocol steps so that individual steps contain only 2-3 actions per step and a maximum of 4 sentences per step. Use sub-steps as necessary.

We have revised the text as recommended.

19. Please add more details to your protocol steps. There should be enough detail in each step to supplement the actions seen in the video so that viewers can easily replicate the protocol. Please ensure you answer the “how” question, i.e., how is the step performed? Alternatively, add references to published material specifying how to perform the protocol action. See examples below.
20. Line 121, 6.4.5: How large is the incision?

We have included the information.

21. Please specify all surgical tools used throughout the protocol.

We have included all the information in text.

22. 3.11: Do you mean “3.5-3.10” instead of “3.5-3.12”?

We have revised the text.

23. 3.12: Please specify the primers and conditions used for genotyping-PCR.

We have included all the information in table.

24. 5.1: Please mention the conditions for growing these cells.

We have revised the text.

25. 5.2: Please describe how the treatment is actually done.

We have revised the text.

26. 5.3: Please specify how cells are harvested.

The steps have been included in 5.4 and 5.5.

27. 6.2.1: Please specify the disinfectant used in this step.

We have revised the text.

28. 6.2.2: Please mention how proper anesthetization is confirmed.

We have revised the text.

29. Please include single-line spaces between all paragraphs, headings, steps, etc.

We have revised the text.

30. After you have made all the recommended changes to your protocol (listed above), please highlight 2.75 pages or less of the Protocol (including headings and spacing) that identifies the essential steps of the protocol for the video, i.e., the steps that should be visualized to tell the most cohesive story of the Protocol.

We have highlighted the text for video filming.

31. Please highlight complete sentences (not parts of sentences). Please ensure that the highlighted part of the step includes at least one action that is written in imperative tense. Notes cannot usually be filmed and should be excluded from the highlighting. Please do not highlight any steps describing anesthetization and euthanasia.

We have highlighted the text for video filming.

32. Please include all relevant details that are required to perform the step in the highlighting. For example: If step 2.5 is highlighted for filming and the details of how to perform the step are given in steps 2.5.1 and 2.5.2, then the sub-steps where the details are provided must be highlighted.

We have revised the text.

33. Discussion: As we are a methods journal, please also discuss critical steps within the protocol, any modifications and troubleshooting of the technique, and any limitations of the technique.

We have revised the text.

34. Please include a Disclosures section, providing information regarding the authors' competing financial interests or other conflicts of interest. If authors have no competing financial interests, then a statement indicating no competing financial interests must be included.

We have included the disclosure statement.

35. References: Please do not abbreviate journal titles.

We have corrected the references